

SEQ ID No.1

atg agt aaa gga gaa gaa ctt ttc act gga gtt gtc cca att ctt gtt	48
gaa tta gat ggt gat gtt aat ggg cac aaa ttt tct gtc agt gga gag	96
ggt gaa ggt gat gca aca tac gga aaa ctt acc ctt aaa ttt att tgc	144
act act gga aaa cta cct gtt cca tgg cca aca ctt gtc act act ttc	192
tct tat ggt gtt caa tgc ttt tca aga tac cca gat cat atg aaa cgg	240
cat gac ttt ttc aag agt gcc atg ccc gaa ggt tat gta cag gaa aga	288
act ata ttt ttc aaa gat gac ggg aac tac aag aca cgt gct gaa gtc	336
aag ttt gaa ggt gat acc ctt gtt aat aga atc gag tta aaa ggt att	384
gat ttt aaa gaa gat gga aac att ctt gga cac aaa ttg gaa tac aac	432
tat aac tca cac aat gta tac atc atg gca gac aaa caa aag aat gga	480
atc aaa gtt aac ttc aaa att aga cac aac att gaa gat gga agc gtt	528
caa cta gca gac cat tat caa caa aat act cca att ggc gat ggc cct	576
gtc ctt tta cca gac aac cat tac ctg tcc aca caa tct gcc ctt tcg	624
aaa gat ccc aac gaa aag aga gac cac atg gtc ctt ctt gag ttt gta	672
aca gct gct ggg att aca cat ggc atg gat gaa cta tac aaa tag	717

[illegible]

Amino Acid Sequence of wtGFP (Chalfie et al, Science, (1994), 263, 802-5

SEQ ID No.2

Met 1	Ser	Lys	Gly	Glu 5	Glu	Leu	Phe	Thr	Gly 10	Val	Val	Pro	Ile	Leu 15	Val
Glu	Leu	Asp	Gly 20	Asp	Val	Asn	Gly	His 25	Lys	Phe	Ser	Val	Ser	Gly	Glu
Gly	Glu	Gly 35	Asp	Ala	Thr	Tyr	Gly 40	Lys	Leu	Thr	Leu	Lys 45	Phe	Ile	Cys
Thr	Thr 50	Gly	Lys	Leu	Pro	Val 55	Pro	Trp	Pro	Thr	Leu 60	Val	Thr	Thr	Phe
Ser 65	Tyr	Gly	Val	Gln	Cys 70	Phe	Ser	Arg	Tyr	Pro	Asp	His	Met	Lys	Arg 80
His	Asp	Phe	Phe 85	Lys	Ser	Ala	Met	Pro	Glu 90	Gly	Tyr	Val	Gln	Glu	Arg 95
Thr	Ile	Phe	Phe 100	Lys	Asp	Asp	Gly	Asn 105	Tyr	Lys	Thr	Arg	Ala	Glu	Val 110
Lys	Phe	Glu 115	Gly	Asp	Thr	Leu	Val 120	Asn	Arg	Ile	Glu	Leu 125	Lys	Gly	Ile
Asp	Phe 130	Lys	Glu	Asp	Gly	Asn 135	Ile	Leu	Gly	His	Lys 140	Leu	Glu	Tyr	Asn
Tyr 145	Asn	Ser	His	Asn 150	Val	Tyr	Ile	Met	Ala	Asp 155	Lys	Gln	Lys	Asn	Gly 160
Ile	Lys	Val	Asn 165	Phe	Lys	Ile	Arg	His	Asn 170	Ile	Glu	Asp	Gly	Ser	Val 175
Gln	Leu	Ala	Asp 180	His	Tyr	Gln	Gln	Asn 185	Thr	Pro	Ile	Gly	Asp	Gly	Pro 190
Val	Leu 195	Leu	Pro	Asp	Asn	His	Tyr 200	Leu	Ser	Thr	Gln	Ser 205	Ala	Leu	Ser
Lys	Asp 210	Pro	Asn	Glu	Lys	Arg 215	Asp	His	Met	Val	Leu 220	Leu	Glu	Phe	Val
Thr 225	Ala	Ala	Gly	Ile 230	Thr	His	Gly	Met	Asp	Glu	Leu 235	Tyr	Lys		

# Figure 3/7

## Predicted Amino Acid Sequence of F64L-S175G-E222G-GFP:

### SEQ ID No.3

Met Ser Lys Gly Glu Glu Leu Phe Thr Gly Val Val Pro Ile Leu Val  
1 5 10 15  
Glu Leu Asp Gly Asp Val Asn Gly His Lys Phe Ser Val Ser Gly Glu  
20 25 30  
Gly Glu Gly Asp Ala Thr Tyr Gly Lys Leu Thr Leu Lys Phe Ile Cys  
35 40 45  
Thr Thr Gly Lys Leu Pro Val Pro Trp Pro Thr Leu Val Thr Thr Leu  
50 55 60  
Ser Tyr Gly Val Gln Cys Phe Ser Arg Tyr Pro Asp His Met Lys Arg  
65 70 75 80  
His Asp Phe Phe Lys Ser Ala Met Pro Glu Gly Tyr Val Gln Glu Arg  
85 90 95  
Thr Ile Phe Phe Lys Asp Asp Gly Asn Tyr Lys Thr Arg Ala Glu Val  
100 105 110  
Lys Phe Glu Gly Asp Thr Leu Val Asn Arg Ile Glu Leu Lys Gly Ile  
115 120 125  
Asp Phe Lys Glu Asp Gly Asn Ile Leu Gly His Lys Leu Glu Tyr Asn  
130 135 140  
Tyr Asn Ser His Asn Val Tyr Ile Met Ala Asp Lys Gln Lys Asn Gly  
145 150 155 160  
Ile Lys Val Asn Phe Lys Ile Arg His Asn Ile Glu Asp Gly Gly Val  
165 170 175  
Gln Leu Ala Asp His Tyr Gln Gln Asn Thr Pro Ile Gly Asp Gly Pro  
180 185 190  
Val Leu Leu Pro Asp Asn His Tyr Leu Ser Thr Gln Ser Ala Leu Ser  
195 200 205  
Lys Asp Pro Asn Glu Lys Arg Asp His Met Val Leu Leu Gly Phe Val  
210 215 220  
Thr Ala Ala Gly Ile Thr His Gly Met Asp Glu Leu Tyr Lys  
225 230 235

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SEQ ID No.4

Met	Ser	Lys	Gly	Glu	Glu	Leu	Phe	Thr	Gly	Val	Val	Pro	Ile	Leu	Val
1				5					10					15	
Glu	Leu	Asp	Gly	Asp	Val	Asn	Gly	His	Lys	Phe	Ser	Val	Ser	Gly	Glu
			20					25					30		
Gly	Glu	Gly	Asp	Ala	Thr	Tyr	Gly	Lys	Leu	Thr	Leu	Lys	Phe	Ile	Cys
		35					40					45			
Thr	Thr	Gly	Lys	Leu	Pro	Val	Pro	Trp	Pro	Thr	Leu	Val	Thr	Thr	Leu
	50					55					60				
Thr	Tyr	Gly	Val	Gln	Cys	Phe	Ser	Arg	Tyr	Pro	Asp	His	Met	Lys	Arg
65					70					75					80
His	Asp	Phe	Phe	Lys	Ser	Ala	Met	Pro	Glu	Gly	Tyr	Val	Gln	Glu	Arg
				85					90					95	
Thr	Ile	Phe	Phe	Lys	Asp	Asp	Gly	Asn	Tyr	Lys	Thr	Arg	Ala	Glu	Val
			100					105					110		
Lys	Phe	Glu	Gly	Asp	Thr	Leu	Val	Asn	Arg	Ile	Glu	Leu	Lys	Gly	Ile
		115					120					125			
Asp	Phe	Lys	Glu	Asp	Gly	Asn	Ile	Leu	Gly	His	Lys	Leu	Glu	Tyr	Asn
	130					135					140				
Tyr	Asn	Ser	His	Asn	Val	Tyr	Ile	Met	Ala	Asp	Lys	Gln	Lys	Asn	Gly
145					150					155					160
Ile	Lys	Val	Asn	Phe	Lys	Ile	Arg	His	Asn	Ile	Glu	Asp	Gly	Gly	Val
				165					170					175	
Gln	Leu	Ala	Asp	His	Tyr	Gln	Gln	Asn	Thr	Pro	Ile	Gly	Asp	Gly	Pro
			180					185					190		
Val	Leu	Leu	Pro	Asp	Asn	His	Tyr	Leu	Ser	Thr	Gln	Ser	Ala	Leu	Ser
	195						200					205			
Lys	Asp	Pro	Asn	Glu	Lys	Arg	Asp	His	Met	Val	Leu	Leu	Glu	Phe	Val
	210					215					220				
Thr	Ala	Ala	Gly	Ile	Thr	His	Gly	Met	Asp	Glu	Leu	Tyr	Lys		
225					230					235					

Figure 5/7

Flow cytometry of GFP mutations

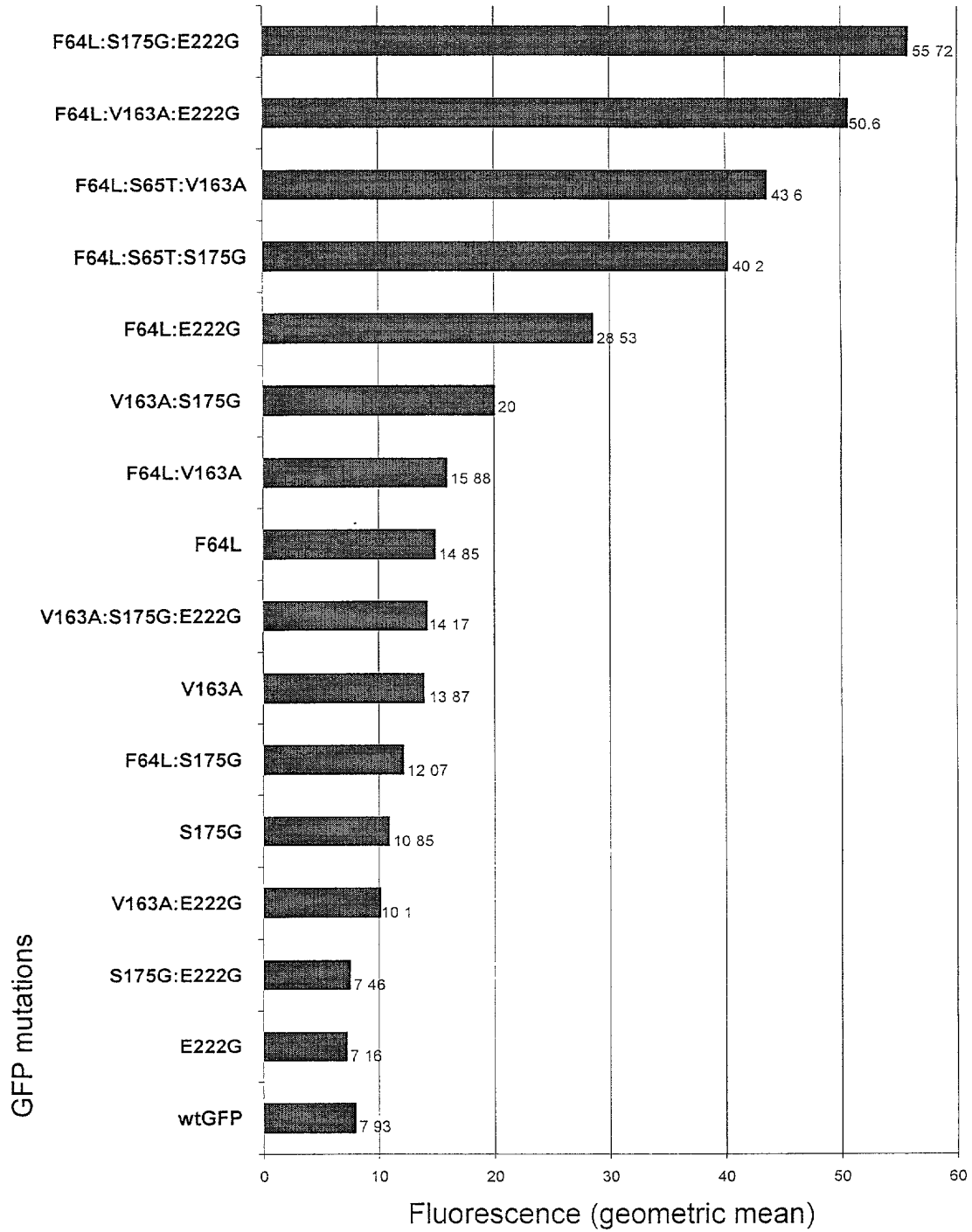


Figure 6/7

Photobleaching of GFP mutations

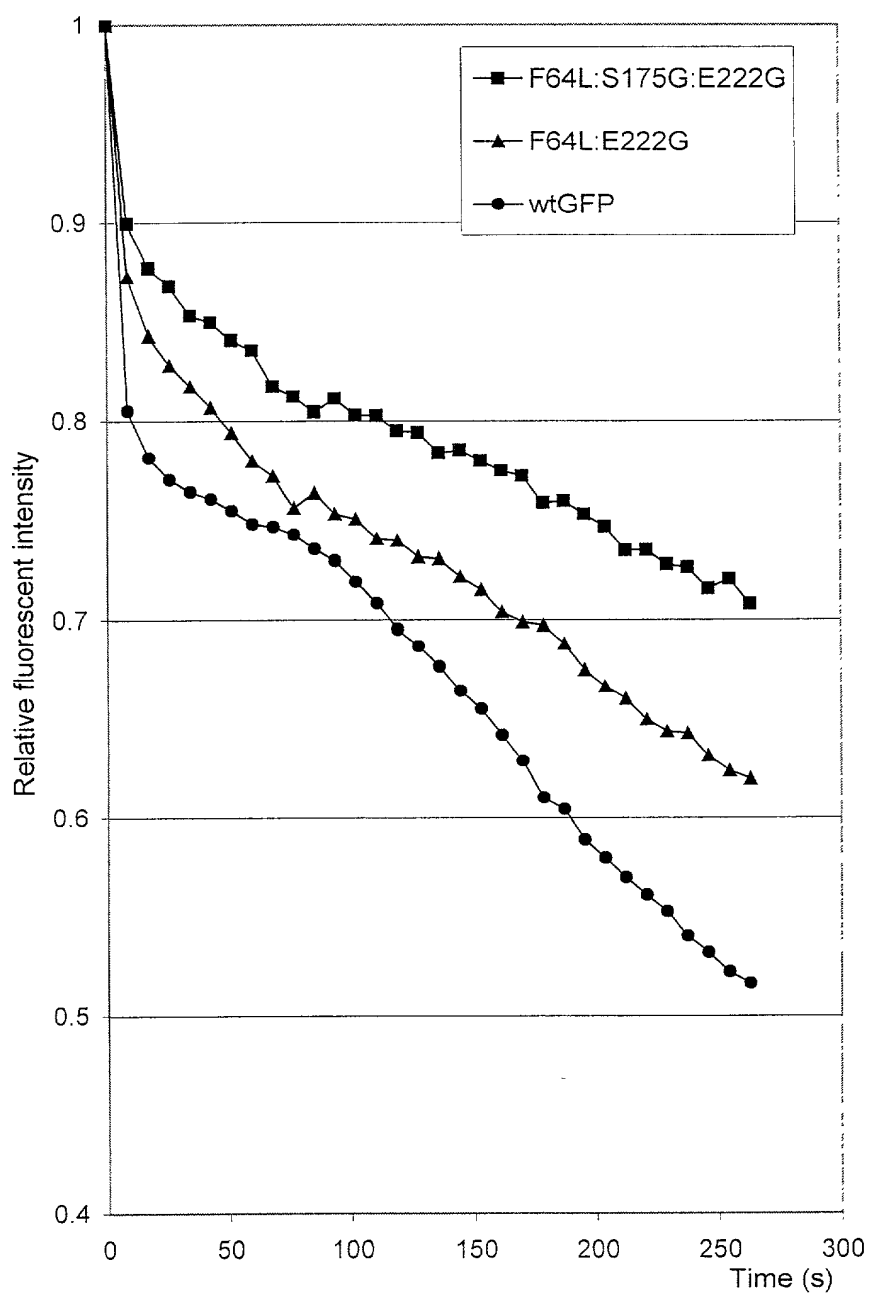


Figure 7/7

Live Cell CHO-HIR NF $\kappa$ B Assay  
P65-tri GFP Assay T = 30 mins

